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High Prevalence of GB-C/Hepatitis G Virus in a Brazilian Population With Helminth Infection

P. Gallian,[1] V. Rodrigues,[2] J.F. Cantaloube,[1] H. Dessein,[2] P. de Micco,[1] A.J. Dessein,[2] and X. de Lamballerie[1]*

[1]*Laboratoire de Virologie Moléculaire Tropicale et Transfusionnelle, Etablissement de Transfusion Sanguine Alpes-Provence, Institut Fédératif de Recherche "Pathologie Transmissible et Pathologie Infectieuse Tropicale," Université de la Méditerranée, Marseille, France*

[2]*INSERM U.399, Immunologie et Génétique des Maladies Parasitaires, Laboratoire de Parasitologie-Mycologie, Institut Fédératif de Recherche "Pathologie Transmissible et Pathologie Infectieuse Tropicale," Université de la Méditerranée, Marseille, France*

A study of GB-C virus/Hepatitis G virus (GBV-C/HGV) infection was carried out in a rural population of Northeastern Brazil, in which the prevalence of schistosomiasis is 80–90%. Despite the absence of parenteral risk exposure, the prevalence of GBV-C/HGV markers of infection was found to be unusually increased: viremia, 16.4%; specific antibody, 18.3%. It is therefore suspected that helminth infection influenced the immune response to GBV-C/HGV infection by shifting the balance of cytokine responses from Th1 to Th2, resulting in a delayed viral clearance. Phylogenetic analysis of viral isolates did not provide evidence for high rates of sexual or mother-to-infant viral transmission. The study revealed that viral strains belonged to types 1 and 2 only (predominant in Africa and Europe, respectively), suggesting that GBV-C/HGV was introduced into the New World by white conquerors and black slaves since the 16th century. ***J. Med. Virol. 56:310–315, 1998.***

KEY WORDS: GBV-C/HGV; schistosomiasis; RT-PCR; anti-E2 antibody; familial distribution

INTRODUCTION

Recently, studies searching for new putative viral agents of non-A–E hepatitis have led to the discovery of several viruses belonging to the *Flaviviridae* family and distantly related to the hepatitis C virus (HCV) [Ohba et al., 1996]. Simons et al. [1995] discovered a new agent called GBV-C in the serum of a West African patient. The following year, Linnen et al. [1996] identified a virus in the plasma of a North American patient with chronic hepatitis and named it hepatitis G virus (HGV). Sequence analysis demonstrated that GBV-C and HGV were two different isolates of the same virus [Zuckerman, 1996], named in this paper "GBV-C/HGV." The different viral strains can be grouped within phylogenetic trees according to their geographical origin [Smith et al., 1997]: isolates of type 1 are closely related to the GBV-C strain isolated in Africa; most isolates from North America and Europe are classified within type 2 that includes the HGV prototype; type 3 has been more recently described and includes essentially Asian isolates.

The virus can be transmitted by blood products [Linnen et al., 1996] and a high prevalence of viral RNA has been reported in populations with parenteral exposure risk, such as multitransfused patients [Sampietro et al., 1997b], hemodialysis patients [de Lamballerie et al., 1996], hemophiliacs [Gonzalez-Perez et al., 1997], and intravenous drug addicts [Diamantis et al., 1997]. However, this mechanism of infection is not the only route that could account for the high prevalence of the biological markers of infection in epidemiological groups with moderate parenteral risks [Nubling et al., 1997] or even in the population of healthy blood donors. Other routes of transmission have been suspected: nosocomial patient-to-patient transmission in hemodialysis units has been suggested [Sampietro et al., 1997a] but has not been formally demonstrated; the importance of sexual [Rubio et al., 1997] or vertical [Fischler et al., 1997] transmission is still unclear.

Similarly, the clinical impact of GBV-C/HGV infection remains uncertain. The virus is able to chronically infect a large proportion of individuals, but this infection is clinically benign in the majority of cases. It has a mild capacity to induce liver damage [Alter, 1997] and does not influence the severity of liver disease or response to interferon alpha therapy in case of coinfection by the hepatitis C virus [Martinot et al., 1997].

*Correspondence to: X. de Lamballerie, Laboratoire de Virologie Moléculaire Tropicale et Transfusionnelle, Faculté de Médecine, Université de la Méditerranée, Bd. J. Moulin, 13385 Marseille Cedex 05, France. E-mail: virophdm@lac.gulliver.fr

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However, the influence of chronic infectious diseases on the natural history of viral infection has not been investigated yet. We report a study in a Brazilian population with high prevalence of schistosome infections and no parenteral risk of viral transmission. We evaluated the prevalence and familial distribution of GBV-C/HGV infection and explored routes of transmission of this agent, using molecular characterization of viral strains.

MATERIALS AND METHODS

Population and Collection of Biological Samples

This study was performed on a randomized sample of 213 persons (100 males and 113 females; mean age, 30; range, 6–85) living in Caatinga do Moura, a village located in a rural area of the Northeast of Brazil (Bahia State). This village is endemic for *Schistosoma mansoni* infection: over the past 10 years, 80–90% of study subjects were found to be infected by this parasite. Accordingly, this population was previously included in a study that permitted the localization of a human locus controlling the intensity of the infection by *Schistosoma mansoni* [Marquet et al., 1996]. Intestinal parasites such as *Ascaris lumbricoides, Strongiloides stercolaris,* and *Entamoeba coli* were observed [Dessein et al., 1984]. Conversely, malaria, leishmaniasis and Chagas' desease were not found in this area. Subjects were treated regularly with antischistosome drugs. However, most of them became reinfected within 1–3 years after treatment.

Blood samples were collected in 1996 by intravenous puncture, and centrifuged at 4,000*g* at 4°C for 15 minutes. Serum was aliquoted and stored frozen at −80°C until use.

Transaminases and Serological Markers of Viral Infection

Alanine aminotransferase (ALT) levels were examined (Biotrol ALT/TGP Bireactif, Merck, Nogent, France), according to the manufacturer's recommendations. Elevated ALT levels were defined by values greater than 1.5 times the value of normal range (men, 6–45 UI/l; women, 5–35 UI/l; <20 years old, 5–25 UI/l), and sub-normal level by values ranging between normal value and 1.5 times the normal value.

Antibodies to hepatitis B core and to HCV and HIV antigens were detected with commercial ELISA kits (HBc ELISA Test System, HVC Third Generation ELISA Test, and HIV-1/HIV-2 Ab-capture ELISA Test System, respectively, Ortho Diagnostic Systems, Raritan, NJ). The detection of antibodies to the envelope protein (anti-E2) of GBV-C/HGV was carried out using the ELISA Enzymun test Anti HGenv (Boehringer Mannheim, Mannheim, Germany).

Detection and Analysis of GBV-C/HGV Sequences

All samples were tested by RT-PCR assay. Extraction of viral RNA and reverse transcription using random hexanucleotides were conducted as described elsewhere [Cantaloube et al., 1997]. The PCR amplification and detection of the 5′ UTR of the GBV-C/HGV genome were performed as reported previously [Cantaloube et al., 1997], using a procedure that was validated by a French multicenter quality control study [Bogard et al., 1997].

Sequence alignments were generated by the Clustal software program (Metrowerks, Inc., Austin, TX). Phylogenetic analysis of 5′-noncoding region sequences was undertaken with the help of the software program MEGA [Kumar et al., 1993], using the Jukes-Cantor algorithm for distance determination and the neighbor-joining method for tree-drawing.

Familial Distribution

The distribution of cases of GBV-C/HGV infection in the different families was studied, using the genealogical trees built for a previous study [Marquet et al., 1996]. Inside each family, the genetic variability of the GBV-C/HGV isolates was studied by comparison of nucleotide sequences of the 5′UTR of the virus.

Statistical Analysis

Evaluation of frequencies between groups using the χ^2 test and comparison of group means using the Mann-Whitney test were undertaken with the help of the SYSTAT software program (SYSTAT, Inc., Evanston, IL). The population was divided in quintiles to constitute age groups including at least 34 individuals.

RESULTS

Markers of Viral Infection

GBV-C/HGV RNA in serum was found in 35 (16.4%) of 213 persons. In the same population, 39 individuals (18.3%) had specific antibodies against the viral E2 envelope glycoprotein. Four persons (2%) were found with GBV-C/HGV RNA and antibodies. Thus, 68 persons (32%) were found to have markers of infection with the virus (Table I).

When the serological status of these individuals against hepatitis B and C viruses and HIV was investigated, it was found that 14 (6.6%) had antibodies to HBV core protein, providing evidence of previous contact with this virus. This result is consistent with previous data showing that HBV infection is endemic in the population of South America [Echevarria et al., 1996]. The absence of parenteral exposure to HCV and HIV in this population was confirmed by the absence of specific antibodies. No case of elevated serum ALT

TABLE I. Distribution of GBV-C/HGV Biological Markers in the Studied Population

	Males, n = 100 (%)	Females, n = 113 (%)	Total n = 213 (%)
GBV-C/HGV RNA	17 (17)	18 (15.9)	35 (16.4)
GBV-C/HGV Ab	19 (19)	20 (17.7)	39 (18.3)
GBV-C/HGV RNA and Ab	0 (0)	4 (3.5)	4 (1.9)

level was detected; only 6 persons had subnormal values, 4 of which had positive HBV markers.

Statistical analysis of these data did not show a significant relation between the presence of GBV-C/HGV markers (RNA or specific antibodies) and sex, level of ALT, or presence of HBV, HCV, and HIV serological markers.

Age and Markers of Infection

The frequency distributions for viremia and specific antibodies in age groups are shown in Figure 1. The most important prevalence for both markers was observed in the group 35–50 years old. Before age 30, the rate of positive PCR was found to be as twice as high as that of antibody-positive samples; beyond this age, the situation was reversed.

The mean age (43.1 years) of the population with antibodies to GBV-C/HGV E2 protein was found to be higher ($P < 0.05$) than that of the population with GBV-C/HGV RNA (34.3 years).

Molecular Analysis of Viral Strains

Thirty-five PCR products of the 5′ UTR were sequenced. When these sequences were aligned with prototype sequences of GBV-C and HGV, homologies with GBV-C and HGV were 81.7–96.7% and 82.5–98.9%, respectively. Brazilian isolates were thus distributed in both type 1 and type 2 groups, according to the classification of Muerhoff et al. [1996]. Figure 2 shows a phylogenetic tree including our 35 Brazilian isolates and GBV-C/HGV isolates from various countries. This evolutionary tree shows that 29 Brazilian GBV-C/HGV isolates were located in the same phylogenetic group as HGV (type 2) that includes a majority of strains from North America and Europe. Six Brazilian isolates were in the GBV-C group (type 1), including mainly strains from West Africa. None of the Brazilian isolates were within the third group (type 3) that encompasses Asian isolates.

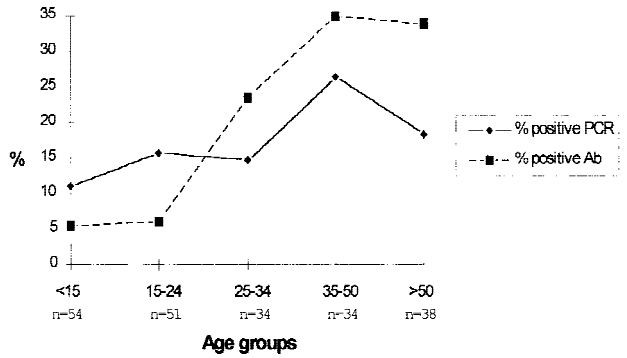

Fig. 1. Distribution of GBV-C/HGV biological markers of infection in age groups. Number of cases and percentage of population in each age group are displayed for the two markers of infection: PCR and specific anti-E2 antibodies (Ab). Results are expressed as percentage in each age group (n = total number).

Familial Distribution of GBV-C/HGV Infection

In 3 cases, it was possible to analyze the viral strains infecting different generations of the same family. A woman, her daughter, and her grandson were found to be PCR-positive for GBV-C/HGV RNA. The three viral strains (Brazil 34, 29, and 2, respectively, indicated by "∗" in Fig. 2) were sequenced in the 5′ UTR of the genome, and were found to be located in three different clusters of the type 2 branch. A similar situation was encountered in another family (mother, strain 24; children, strains 9, 12, 17, and 3; indicated by "⋆" in Fig. 2). In a third family, the mother's viral strain belonged to type 2b, while her son's strain was characterized as a type 1 isolate (strains Brazil 8 and 30, respectively, indicated by "+" in Fig. 2). Thus in all cases, there was no evidence for a mother-to-infant transmission of the virus.

In the case of 8 married couples, the status of both wife and husband could be studied. In 3 couples, the man and the woman had biological markers of the infection. In the case of each of 5 spouses (all with several children), one person had a positive marker (viremia, n = 3; specific antibody, n = 2), but the other had none. This did not suggest efficiency of sexual transmission of the virus in this population.

DISCUSSION

Several studies have been conducted in various populations to determine the prevalence of GBV-C/HGV infection. These clearly demonstrated that injection of blood products increased the risk of infection. The Brazilian population that we studied clearly had no risk of parenteral exposure. No case of HCV or HIV infection was found, and previous studies had shown that this population had no access to blood transfusion, immunoglobulins, and all other components of parenteral therapy. The intravenous use of drugs was absent, and there is no tradition of rituals such as scarification or circumcision. In this context, the high prevalence of markers of contact with GBV-C/HGV (viremia, 16.4%; antibodies, 18.3%) cannot be explained by parenteral transmission of this virus. This problem had already been seen in populations of blood donors without any identifiable risk factor, in which a prevalence in the order of 15% was found [Cantaloube, personal data; Feucht et al., 1997; Nubling et al., 1997]. It is interesting to note, as discussed below, that the positive serology/positive PCR ratio ranges between 5–7 in such populations, and is only 1 in this study.

No evidence was found in this population for a relation between the GBV-C/HGV infection and a particular pathological syndrome. Transaminases were not more elevated in the group of individuals with a positive PCR than in the rest of the population. There was no relation between the markers of infection and the sex, or the presence of antibodies to the hepatitis B and C viruses. The mean age of the individuals with a positive PCR (34.3 years old) was found to be lower than that of individuals with specific antibody to GBV-C/HGV (43.1 years old). The frequency distributions in

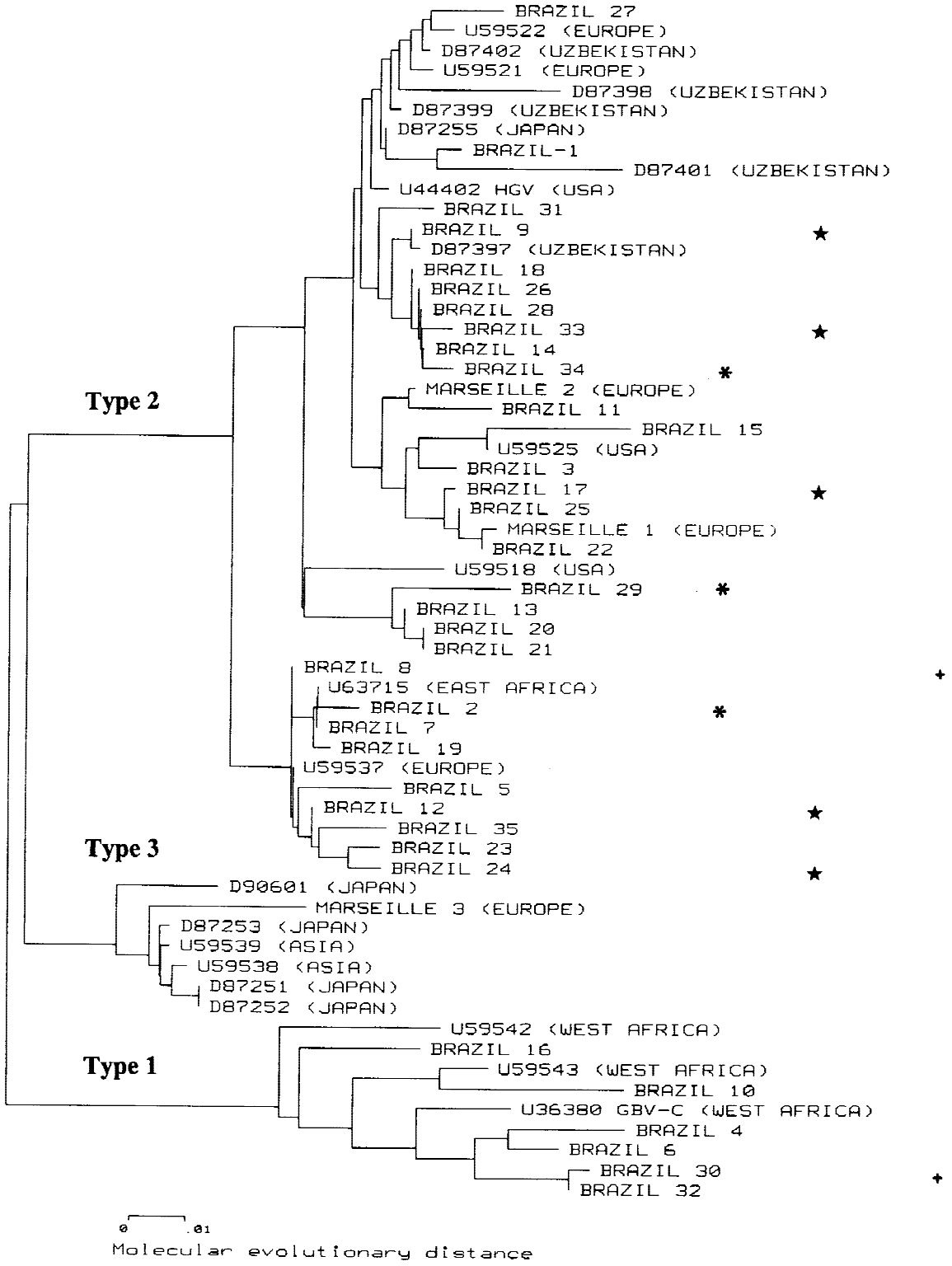

Fig. 2. Unrooted phylogenetic tree from GBV-C/HGV 5′ UTR sequences alignment, including isolates collected from 35 Brazilian individuals (strains Brazil 1–35), from 3 blood donors of Marseille (Marseille 1–3) and 22 prototype sequences collected from the GenBank database (accession numbers U44402, U63715, U36380, D90601, D87397–D87399, D87401, D87402, U59518, U59521, U59522, U59525, U59537–U59539, U59542, U59543, D87251–D87253, and D87255). Identification of strains ∗, ⋆, and + refers to the identification of 3 families, as detailed in Results.

age groups showed that the proportion of positive PCR was higher than that of positive serological findings in young individuals, and that this situation was reversed after age 30. This suggests that a large proportion of infected patients finally produces antibodies to the virus and recovers from infection.

We analyzed the distribution of biological markers within different families, in order to try to elucidate the routes of infection in this population. One possibility was sexual transmission [Kao et al., 1997]. We examined 8 couples (all with several children) and found that in 5 cases, one person had a biological marker of infection and the other had none. This obviously does not indicate sexual transmission of the infection in this population. In 3 cases, the possibility of a vertical transmission of GBV-C/HGV was investigated, using molecular tools. In all cases, it was shown that different strains were infecting the child and mother. Consequently, it is concluded that vertical transmission of the virus is not frequent. These findings are consistent with those of Dawson et al. [1996], who found that vertical transmission could not be the sole route of transmission in a Ghanaian population, since the prevalence rates increased with age.

In the study area, helminth infection is endemic, and 80–90% of the population are infected with *Schistosoma mansoni.* This human parasitic infection has been shown to be frequently associated with hepatitis B infection [Bassily et al., 1983], and in a recent study GBV-C/HGV viremia was found to be significantly associated with a history of schistosomiasis in Egyptian patients [Hassoba et al., 1997]. The infection by *Schistosoma mansoni* is known to skew in a Th2 direction the immune response to antigens or pathogens that normally induces Th1 responses. Actor et al. [1993] reported that BALB/c mice infected by *S. mansoni* displayed severely impaired CD8+ class I major histocompatibility complex-restricted CTL (Cytotoxic T-lymphocyte) function against a recombinant vaccina virus expressing the HIV-1 gp 160 glycoprotein. As a consequence, the immune clearance of viral infection was severely delayed in parasited animals. The population of Caatinga do Moura was studied by Sabin et al. [1996], and these investigators demonstrated that *S. mansoni*-infected persons mounted a Th2-like response to tetanus toxoid antigen, while uninfected persons mounted a Th1- or Th0-like response. Thus, an immune mechanism similar to that described by Actor et al. [1993] could be responsible for the high prevalence of GBV-C/HGV infection that we observed. The helminth infection could downregulate viral-specific CTL responses and shift the balance in T-helper cytokine responses from Th1 to Th2. This would result in a prolonged GBV-C/HGV viremia and could explain the unusually low positive serology/positive PCR ratio in this population.

The phylogenetic study of the viral strains encountered in this population identified isolates from the 1a, 1b, 2a, and 2b subtypes. The mean age of the individuals infected by each subtype was found to be similar (30–35 years old) and comparable to that of the sample of infected persons. These data are consistent with the absence of one or several successive outbreaks. They suggest that infection by a large number of different strains is endemic in this population. The distribution of Brazilian strains within a phylogenetic tree did not identify a cluster including specific isolates from South America. We did not observe any strain of type 3, in contrast to a study of Nicaraguan hemodialysis patients [Gonzalez-Perez et al., 1997]. All the isolates belonged to type 1 (including mostly African strains) and 2 (including North American and European strains). This is consistent with the ethnic origin of the Brazilian population, resulting from the crossbreeding of the original Amerindians with European and African populations which migrated to South America since the 16th century. It is therefore probable that GBV-C/HGV was imported to South America by immigrants from Europe and Africa, as previously demonstrated for several infectious agents [Newman, 1976], and that it has been present in Brazil for almost 300 years. Such a hypothesis would imply that the high similarity of the Brazilian strains with the original European and African isolates is due to the very low variability of the virus.

REFERENCES

Actor JK, Shirai M, Kullberg MC, Buller RML, Sher A, Berzofsky JA (1993): Helminth infection results in decreased virus-specific CD8+ cytotoxic T-cell and th1 cytokine responses as well as delayed virus clearance. Proceedings of the National Academy of Sciences of the United States of America 90:948–952.

Alter HJ (1997): G-pers creepers, where'd you get those papers? A reassessment of the literature on the hepatitis G virus. Transfusion 37:569–572.

Bassily S, Dunn MA, Farid Z, Kilpatrick ME, El-Masry NA, Kamel IA, El-Alamy M, Murphy BL (1983): Chronic hepatitis B in patients with Schistosomiasis mansoni. Journal of Tropical Medicine and Hygiene 86:67–71.

Bogard M, Buffet-Janvresse C, Cantaloube JF, Biagini P, Duverlie G, Castelain S, Izopet J, Dubois M, Defer C, Lepot I, Coste J, Marcellin P, Martinot-Peignoux M, Halfon P, Gerolami V, Frangeul L, Pawlotsky JM, Roudot-Thoraval F, Dussaix E, Loiseau P, Ravera N, Lewin P, Lamoril J, Lerable J, Bouchardeau F, Laperche S, Mariotti M, Lefrère JJ, Nicot T, Denis F, Gassain M, Mérel P, Trépo C, Thiers V, Bréchot C, Férec C, Mercier B, Stoll-Keller F, Palmer P, Lebon P (1997): GEMHEP multicenter quality control study of PCR detection of GB virus C/hepatitis G virus RNA in serum. Journal of Clinical Microbiology 35:3298–3300.

Cantaloube JF, Charrel RN, Attoui H, Biagini P, De Micco P, de Lamballerie X (1997): Evaluation of four PCR systems amplifying different genomic regions for molecular diagnosis of GB virus C infections. Journal of Virological Methods 64:131–135.

Dawson GJ, Schlauder GG, Pilot-Matias TJ, Thiele D, Leary TP, Murphy P, Rosenblatt JE, Simons JN, Martinson FEA, Gutierrez RA, Lentino JR, Pachucki C, Muerhoff AS, Widell A, Tegtmeier G, Desai SM, Mushahwar IK (1996): Prevalence studies of GB virus-C infection using reverse transcriptase-polymerase chain reaction. Journal of Medical Virology 50:97–103.

de Lamballerie X, Charrel RN, Dussol B (1996): Hepatitis GB virus C in patients on hemodialysis. New England Journal of Medicine 334:1549.

Dessein AJ, Lenzi HL, Bina JC, Carvalho EM, Weiser WY, Andrade ZA, David JR (1984): Modulation of eosinophil cytotoxicity by blood mononuclear cells from healthy subjects and patients with chronic schistosomiasis mansoni. Cellular Immunology 85:100–113.

Diamantis I, Bassetti S, Erb P, Ladewig D, Gyr K, Battegay M (1997): High prevalence and coinfection rate of hepatitis G and C infections in intravenous drug addicts. Journal of Hepatology 26:794–797.

Echevarria JM, Blitz-Dorfman L, Pujot FH (1996): Infection by hepatitis virus among the indigenous populations of South America: A review of the problem. Investigacion Clinica 37:191–200.

Feucht HH, Zollner B, Polywka S, Knodler B (1997): Distribution of hepatitis G viremia and antibody response to recombinant proteins with special regard to risk factors in 709 patients. Hepatology 26:491–494.

Fischler B, Lara C, Chen M, Sonnerborg A, Nemeth A, Sallberg M (1997): Genetic evidence for mother-to-infant transmission of hepatitis G virus. Journal of Infectious Diseases 176:281–285.

Gonzalez-Perez MA, Norder H, Bergstrom A, Lopez E, Visona KA, Magnius LO (1997): High prevalence of GB virus C strains genetically related to strains with Asian origin in Nicaraguan hemophiliacs. Journal of Medical Virology 52:149–155.

Hassoba HM, Terrault NA, El-Gohary AM, Scheffel C, Jou C, Brackett J, Hunt J, Chang Lou S, Wright TL (1997): Antibody to GBV-C second envelope glycoprotein (anti GBV-C E2): Is it a marker for immunity? Journal of Medical Virology 53:354–360.

Kao JH, Liu CJ, Chen PJ, Chen W, Hsiang SC, Lai MY, Chen DS (1997): Interspousal transmission of GBVirus-C/Hepatitis G Virus: A comparison with hepatitis C Virus. Journal of Medical Virology 53:348–353.

Kumar S, Tamura K, Nei M (1993): "Molecular Evoluationary Genetics Analysis, Version 1.01." University Park, PA: Pennsylvania State University.

Linnen J, Wages J, Zhang-Keck ZY, Fry KE, Krawczynski KZ, Alter H, Koonin E, Gallagher M, Alter M, Hadziyannis S, Karayiannis P, Fung K, Nakatsuji Y, Shih JWK, Young L, Piatak M, Hoover C, Fernandez J, Chen S, Zou JC, Morris T, Hyams KC, Ismay S, Lifson JD, Hess G, Foung SKH, Thomas H, Bradley D, Margolis H, Kim JP (1996): Molecular cloning and disease association of hepatitis G virus: A transfusion-transmissible agent. Science 271:505–508.

Marquet S, Abel L, Hillaire D, Dessein H, Kalil J, Feingol, Weissenbach J, Dessein AJ (1996): Genetic localization of a locus controlling the intensity of infection by *Schistosoma mansoni* on chromosome 5q31–q33. Nature Genetics 14:181–184.

Martinot M, Marcellin P, Boyer N, Detmer J, Pouteau M, Castelnau C, Degott C, Auperin A, Collins M, Kolberg J, Wilber J, Benhamou JP, Erlinger S (1997): Influence of hepatitis G virus infection on the severity of liver disease and response to interferon-alpha in patients with chronic hepatitis C. Annals of Internal Medicine 126:874–881.

Muerhoff AS, Simons JN, Leary TP, Erker JC, Chalmers ML, Pilot-Matias TJ, Dawson GJ, Desai SM, Mushahwar IK (1996): Sequence heterogeneity within the 5′-terminal region of the hepatitis GB virus C genome and evidence for genotypes. Journal of Hepatology 25:379–384.

Newman MT (1976): Aboriginal New World epidemiology and medical care, and the impact of Old World disease imports. American Journal of Physical Anthropology 45:667–672.

Nubling CM, Biallek H, Fursch AJ, Scharrer I, Schramm W, Seifried E, Schmidt U, Staszewski S, Lower J (1997): Frequencies of GB virus-C/hepatitis G virus genomes and of specific antibodies in German risk and nonrisk populations. Journal of Medical Virology 53:218–224.

Ohba K, Mizokami M, Lau JY, Orito E, Ikeo K, Gojobori T (1996): Evolutionary relationship of hepatitis C, pesti-, flavi-, plantviruses, and newly discovered GB hepatitis agents. FEBS Letters 378:232–234.

Rubio A, Rey C, Sanchez-Quijano A, Leal M, Pineda JA, Lissen E, Hess G (1997): Is hepatitis G virus transmitted sexually? JAMA 277:532–533.

Sabin EA, Araujo MI, Carvalho EM, Pearce EJ (1996): Impairment of tetanus toxoid-specific Th1-like immune responses in humans infected with Schistosoma mansoni. Journal of Infectious Diseases 173:269–272.

Sampietro M, Badalamenti S, Graziani G, Como G, Buccianti G, Corbetta N, Ticozzi A, Archenti A, Lunghi G, Penso D, Pizzuti A, Fiorelli G, Ponticelli G (1997a): Hepatitis G virus infection in hemodialysis patients. Kidney International 51:348–352.

Sampietro M, Corbetta N, Cerino M, Fabiani P, Ticozzi A, Orlandi A, Lunghi G, Fargion S, Fiorelli G, Cappellini MD (1997b): Prevalence and clinical significance of hepatitis G virus infection in adult beta-thalassaemia major patients. British Journal of Haematology 97:904–907.

Simons JN, Leary TP, Dawson GJ, Pilot-Matias TJ, Muerhoff AS, Schlauder GG, Desai SM, Mushahwar IK (1995): Isolation of novel virus-like sequences associated with human hepatitis. Nature Medecine 1:564–569.

Smith DB, Cuceanu N, Davidson F, Jarvis LM, Mokili JLK, Hamid S, Ludlam CA, Simmonds P (1997): Discrimination of hepatitis G virus/GBV-C geographical variants by analysis of the 5′ noncoding region. Journal of General Virology 78:1533–1542.

Zuckerman AJ (1996): Alphabet of hepatitis viruses. Lancet 347:558–559.